BP 2: Physics of Cells I

Time: Monday 9:30–13:00 Location: H 1028

Invited Talk BP 2.1 Mon 9:30 H 1028

Membrane tension regulates motility by controlling lamellipodium organization — •Julie Plastino — Institut Curie, Paris, France

Many cell movements proceed via a crawling mechanism, where assembly of the cytoskeleton pushes out the leading edge membrane. In this scenario, membrane tension has been seen as an impediment to cytoskeleton polymerization and cell motility. Here we exploit a simple model of cell motility, the Caenorhabditis elegans sperm cell, and we apply osmotic and biochemical treatments that relax or tense the cell membrane, in order to test how membrane tension affects cytoskeleton dynamics and cell movement. Surprisingly, we find that membrane tension reduction is correlated with a decrease in cell displacement speed, whereas an increase in membrane tension enhances motility. We propose and show evidence for the idea that membrane tension optimizes motility by streamlining polymerization in the direction of movement, thus adding a layer of complexity to our current understanding of how membrane tension enters into the motility equation.

BP 2.2 Mon 10:00 H 1028

Mixing dynamics of the actin cytoskeleton in motile cells — •Matthias Gerhardt, Michael Walz, and Carsten Beta — Institut für Physik und Astronomie, Karl-Liebknecht-Strasse 24/25, 14476 Potsdam-Golm, Germany

We investigate the dynamics of the actin cytoskeleton during and immediately after electrofusion of cells of the social amoeba Dictyostelium discoideum. Two cell lines are used, one expressing a green (GFP) the other one a red fluorescent protein (mRFP) linked to a Lim-domain protein that serves as a marker for filamentous actin. Using a custom designed fusion chamber mounted on a confocal microscope, we are able to study the mixing dynamics of the actin cytoskeleton during cell fusion. The fusion process typically proceeds via three stages: I.) The early state is characterized by an initial pore, which connects the cells. F-actin fragments with a green label invade the cell expressing the red label and vica versa. II.) In the intermediate state, the cells are connected by a junction of hour-glass shape. The actin cytoskeleton starts mixing, resulting in a yellow overlay of the two labels. III.) The final state is characterized by a completely mixed actin cytoskeleton in a fused cell. The timescale of mixing and remodeling of the cytoskeleton is surprisingly short, indicating that the active, self-organized processes in the actin cytoskeleton quickly integrate constituents of both cells into one overall cortex of the fused cell.

BP 2.3 Mon 10:15 H 1028

Contractile forces facilitate the enhanced glycosylphosphatidylinositol-anchored receptor CD24-dependent invasiveness of cancer cells — •Claudia Tanja Mierke — University of Leipzig, Institute for Experimental Physics I, Soft Matter Physics Division

The malignancy of tumors depends on the capability of cancer cells to metastasize. The process of metastasis involves cell invasion through the extracellular matrix (ECM). Cell invasion is a fundamental biomechanical process, which usually requires adhesion to the ECM through mainly beta1 heterodimeric integrin receptors. The localization of beta1 integrins to lipid rafts depends on the glycosylphosphatidylinositol-anchored receptor CD24. The expression of CD24 is up-regulated in several tumor types and consistently associated with increased metastasis in patients. Here, the invasion of A125 lung cancer cells with different CD24 expression levels was studied in 3D-ECMs. The hypothesis was that CD24 expression increases the invasion of cancer cells through increased contractile forces. To analyze this, A125 cells (CD24negative) were stably transfected with CD24 and sorted for high and low CD24 expression. The invasiveness of CD24high and CD24low transfectants were determined in 3D-ECMs. The percentage of invasive cells and their invasion depth were increased in CD24high compared to CD24low cells. Fourier-transform-traction-microscopy revealed that CD24high cells generated 5-fold higher contractile forces compared to CD24low cells. Finally, these results suggest that CD24 enhances cell invasion through increased contractile forces.

BP 2.4 Mon 10:30 H 1028

Cell shape and behaviour for accurate chemotaxis — •Luke Tweedy[1,2], Börn Meier[3,4], Jürgen Stephan[3,4], Doris Heinrich[3,4], and Robert Endres[1,2] — [1]Division of Molecular Biosciences, Imperial College, London, United Kingdom — [2]Centre for Integrative Systems Biology and Bioinformatics, London, United Kingdom — [3]Ludwig-Maximilians-University, Munich, Germany — [4]Center for NanoScience, Munich, Germany

The behaviour of an organism often reflects a strategy for coping with its environment. Such behaviour in higher organisms can often be reduced to a few stereotyped modes of movement due to physiological limitations, but finding such modes in amoeboid cells is more difficult as they lack these constraints. Here, we investigate the connection of stereotypical cell shape and movement in the amoeba Dictyostelium discoideum with its ability to accurately chemotax. We show that the incredible variety in amoeboid shape can be reduced to a few principal modes, which capture the majority of variability in the population. The cell*s preference for modes depends on the chemical environment. We further construct a parameter-free model using the principle of maximum caliber, which accurately predicts long-term cell behaviour. Stereotypy in cells may thus inform our understanding of cell physiology, evolution, and strategy, and may even be used to screen cell health.

BP 2.5 Mon 10:45 H 1028

Cytoplasmic streaming in giant algae cells: the role of wall slip — •Katrin Wolff, Davide Marenduzzo, and Mike Cates — SUPA, School of Physics & Astronomy, University of Edinburgh, UK

We present lattice Boltzmann simulations of a microscopic model for cytoplasmic streaming in algal cells such as those of Chara corallina. The fluid motion is driven by myosin motors carrying vesicles or other organelles and moving along actin filaments which are attached to the outer part of the cytoplasm. We address how the high speeds observed in experiments can be achieved by assuming a layer of lower viscosity at the outer wall of the simulated compartment. To this end we introduce a finite slip boundary condition at the wall close to which the motors move. The motivation behind the low-viscosity layer is the assumption that those cell contents populating the cytoplasm do not reach up to the cell wall resulting in a more dilute solution close to the wall. We find that this simplified view, which does not rely on any coupling between motors, cytoplasm and vacuole other than that provided by viscous Stokes flow, accounts very well for the observed magnitude of streaming velocities [1].

[1] K. Wolff, D. Marenduzzo, M. Cates, Cytoplasmic streaming in giant algae cells: the role of wall slip (submitted)

BP 2.6 Mon 11:00 H 1028

Mechanics in Neuronal Development — •Kristian Franze[1,2], Hanno Svoboda[1], Andreas F. Christ[2], Luciano da F. Costa[3], Christine E. Holt[1], and Jochen Guck[2] — [1]Department of Physiology, Development and Neuroscience — [2]Department of Physics, University of Cambridge, UK — [3]Instituto de Fisica de Sao Carlos, University of Sao Paulo, Brazil

During the development of the nervous system, neurons migrate and grow over great distances. During these processes, they are exposed to a multitude of signals influencing their speed and direction. Currently, our understanding of neuronal development is, in large part, based on studies of biochemical signalling. Despite the fact that forces are involved in any kind of active cell motion, mechanical signalling has so far rarely been considered. Here we used atomic force microscopy to study the mechanical properties of developing brain tissue. Additionally, we exploited deformable cell culture substrates, traction force microscopy and calcium imaging to investigate how neurons respond to their mechanical environment. The tendency to grow in bundles, which neurons show in vivo, was significantly enhanced on soft substrates. Moreover, if grown on substrates incorporating linear stiffness gradients, neurons were repelled by stiff substrates. Calcium influxes through the activation of stretch-activated ion channels appear to be involved in neuronal mechanosensitivity. A comparison of our in vitro findings with the neurons' in vivo environment suggests that mechanical signalling is involved in neuronal pathfinding and constitutes a formerly unknown guidance mechanism.

15 min break

BP 2.7 Mon 11:30 H 1028

Inherently slow and weak forward forces of neuronal growth cones measured by a drift-stabilized Atomic Force Microscope — •Thomas Fuhs[1], Lydia Reuter[1], Iris Vonderhaid[1], Thomas Claudepierre[2], and Josef A. Käs[1] — [1]Universität Leipzig, Soft matter physics, Leipzig, Germany — [2]Universitätsklinikum Leipzig, Klinik und Poliklinik für Augenheilkunde, Leipzig, Germany

Previous results have convincingly shown that neurons prefer soft environments, such as glia cells. This assures that neurons are confined to the central nervous system and cannot wander off. Nevertheless, the question remains, whether or not growth cones have the ability to migrate in stiffer environments like glial scars, as required in nerve regeneration. We investigated the mechanical properties and force generation of extending retinal ganglion cells and NG108-15 growth cones using different AFM based methods. With our drift-stabilized AFM we could, for the first time, measure the forward pushing forces at the leading edge of outgrowing neuronal growth cones. Our results demonstrate that growth cones have neither the required stability nor the ability to produce forces necessary to penetrate hard tissues.

BP 2.8 Mon 11:45 H 1028

Reconstruction of Cellular Forces During Migration Through Three-Dimensional Collagen Meshworks — •Julian Steinwachs, Claus Metzner, Nadine Lang, Navid Bonakdar, Stefan Münster, and Ben Fabry — Biophysics Group Universität Erlangen-Nürnberg

Reconstituted collagen gels are a widely used substitute of connective tissue to study cell migration in three dimensions. The importance of cellular traction forces needed for the cells to overcome the steric hindrance of the connective tissue is still unknown. We developed a method to quantify cell tractions in a highly nonlinear fibrous biopolymer 3-D network such as collagen. Using confocal reflection microscopy we image the 3-D fiber structure around cells as they migrate through the collagen gels. Cell forces are then relaxed using cytochalasin D, and the relaxed state of the gel is also imaged. Using a fiber pattern matching algorithm, we reconstruct the cell-induced deformation field around every cell. Matrix stresses are computed with a model for the elastic behavior of collagen gels in which the non-affine behavior (buckling, alignment and tautening of fibers) is approximated by a network of nonlinear elements that deform in an affine way. The model parameters are obtained from rheological measurements. We then optimize the cell tractions that best account for the measured deformation field with a least squared optimization routine. If the precise cell contour is unknown, it is still possible to reconstruct the stress field around the cell and to quantify the traction magnitude. Total computation time is less than 5 minutes per cell on an average desktop computer.

BP 2.9 Mon 12:00 H 1028

Traction Force Reconstruction based on Finite Element Methods — •Jérôme Soiné[1,2] and Ulrich Schwarz[1,2] — [1]Bioquant, Heidelberg University, Heidelberg, Germany — [2]Institute for Theoretical Physics, Heidelberg University, Heidelberg, Germany

Over the last decade, it has been established that tissue cells are able to sense the stiffness, geometry and topography of their adhesive environment by actively applying forces to the cell-matrix interface. Cellular forces can be reconstructed with *traction force microscopy*. This method combines cell culture experiments on soft elastic substrates with quantitative image processing and solution of the inverse problem of elasticity theory. Traditional approaches are based on the well-known exact Green's function for planar linear elastic materials known as *Boussinesq solution* [1-3]. For non-planar substrates or non-linear material laws, this approach fails due to the lack of exact solutions. In order to overcome these limitations, we have developed a force reconstruction technique that uses a numeric optimization approach based on finite element methods. It allows us to reconstruct cellular forces for arbitrary substrate geometry and different material laws with a resolution comparable to previous approaches.

[1] Dembo and Wang, Biophysical Journal, 1999
[2] Butler et al., Am. J. Physiol. Cell Physiol., 2002
[3] Sabass et al., Biophysical Journal, 2008

BP 2.10 Mon 12:15 H 1028

Probing PI3-kinase based cell reorientation in spatio-temporally controlled chemotactic gradient fields — Boern Meier and •Doris Heinrich — Center for Nanoscience (CeNS) and Faculty of Physics, Ludwig-Maximilians-Universität München, Geschwister-Scholl-Platz 1, 80539 München

We developed a microfluidic chamber to manipulate cell migration in spatio-temporally controlled gradient fields. Bidirectional chemical gradients over a width of more than 300 um and timescales from seconds to several hours allow for parallel exposure of entire cell ensembles. This setup greatly facilitates statistical analysis of cellular migration properties in response to changing gradient directions and for genetic or pharmacological perturbation of the underlying regulatory network.

The long standing observation that actin polymerization, actuated by the phosphorylation of PIP2 by PI3-kinase, is primarily mediating chemotactic migration, has recently been challenged by the observation of Phospholipase A2 induced pseudopod splitting at the leading edge, which enhances persistence in directed cell migration. In Dictyostelium discoideum cells, we find that PI3-kinase based formation of new pseudopods is promoted by steep chemotactic gradients and reduced cellular starvation times, while reduction of the gradient steepness and ongoing starvation enhances persistent cell migration. Consecutive experiments with increased gradient switching frequencies promise further insight into the dynamic regulation of both parallel feedback loops.

BP 2.11 Mon 12:30 H 1028

Quantitative Investigations of Circular Dorsal Ruffling Dynamics — •Erik Bernitt[1,3], Malte Ohmstede[1], Pritpal Singh[2], Cheng-Gee Koh[2], and Hans-Günther Döbereiner[1,3] — [1]Institut für Biophysik, Universität Bremen, 28334 Bremen — [2]School of Biological Sciences, Nanyang Technological University, 639798 Singapore — [3]Mechanobiology Institute Singapore, National University of Singapore, 117411 Singapore

Circular dorsal ruffles (CDRs) are actin-based structures that, unlike peripheral ruffles, form on the dorsal side of cells. Upon initialization, a membrane sheet of vertical extension forms that propagates in a ring-like fashion over the cell. After formation CDRs enlarge, sometimes covering the complete dorsal side of the cell, propagate and collapse to one point. CDRs are known to occur, e.g., on fibroblast cells upon growth factor stimulation. Even though this has been known for decades, the underlying mechanisms leading to formation and propagation of these coherent solitary waves are not understood. Only recently models have been published that describe CDRs based on diffusion reaction processes in the cytosol and on the membrane.

We use DIC-based optical sectioning in conjunction with image processing for localisation and the three-dimensional reconstruction of CDR morphology. We are the first to describe the CDR dynamics in terms of morphology and propagation velocities in great detail. We correlate these data with fluorenscence data of the actin cytoskeleton, providing the first dynamic data on the onset of the migratory state, one hypothesized biological function of CDRs.

BP 2.12 Mon 12:45 H 1028

Viscosity-Sensing and Mechano-Transduction of Cells on Adhesive Lipid Bilayers — Lena Astrid Lautscham[1], Corey Yu-Hung Lin[2], •Daniel Minner[2], Wolfgang Goldmann[1], Christoph Naumann[2], and Ben Fabry[1] — [1]University Erlangen-Nuremberg, Department of Physics — [2]Indiana University-Purdue University, Department of Chemistry and Chemical Biology

Adherent cells have been shown to sense the mechanical properties of their extracellular matrix and to respond to it by altering their morphology, migration speed, and cytoskeletal organization. Previous studies focused on substrate elasticity to investigate cellular mechano sensing and transduction. Here, we altered the substrate viscosity as an alternative route to probe the influence of matrix mechanics on cell responses. Polymer-tethered multi-lipid bilayer systems on a solid support provide a method to tune the substrate viscosity over an extended range by altering the number of stacked bilayers. In contrast to elastic substrates where deformations come to a halt when cell tractions reach a steady state, cell adhesion ligands in viscous substrates remain mobile and thus provide a different mechanical stimulus. To maintain cell tractions, cells need to continuously reorganize their focal adhesions and associated cytoskeletal structures. We probed mechanical bilayer properties as well as cytoskeletal properties with magnetic tweezers. Our data indicate that mouse embryonic fibroblasts (MEF) are extremely susceptible to decreasing substrate viscosity and respond by altering their cytoskeletal and focal adhesion dynamics, cytoskeletal stiffness and spreading area.